

Claims

1. A method for determining a characteristic kinetic quantity of a chemical reaction in a sample involving a plurality of chemical species, at least one of said species including at least one fluorophore, the method comprising the steps of:

generating, by impinging light on said sample, a non-equilibrium state of said chemical reaction, and

observing, by means of a fluorescence signal of at least one fluorophore, at least one portion of a relaxation of concentrations of said species involved,

the method being characterized in that

at least one product of said chemical reaction under test comprises a combination of two species each of which including one partner of a FRET pair consisting of a FRET donor and a FRET acceptor wherein

said FRET acceptor is a photochrome, the absorption spectrum of which being changeable by irradiation with light of a suitable wavelength;

said FRET donor is a fluorophore, the emission spectrum of which having an overlap region with said FRET acceptor's absorption spectrum, the size of said overlap region being dependent on the photochromic state of said FRET acceptor; and

said light used for generating said non-equilibrium state has a wavelength capable of switching said photochromic state of said FRET acceptor.

2. A method according to claim 1, characterized in that the fluorescence of said FRET donor is measured in order to observe said relaxation.

3. A method according to any of the preceding claims, characterized in that said photochromic FRET acceptor is a fluorophore and that the fluorescence of said photochromic FRET acceptor is measured in order to observe said relaxation.

4. A method according to any of the preceding claims, characterized in that the product under test comprises an additional fluorophore which represents an additional FRET acceptor to said FRET donor.

5. A method according to claim 4, characterized in that said additional FRET acceptor is no photochrome.

6. A method according to any of the claims 4 and 5, characterized in that the fluorescence of said additional FRET acceptor is measured in order to observe said relaxation.

7. A method according to any of the preceding claims, characterized in that a change in the photochromic state of said FRET acceptor in a first direction is caused by irradiation of said sample with light of a first wavelength and that a change in the photochromic state of said FRET acceptor in a second direction is caused by irradiation of said sample with light of a second wavelength.

8. A method according to any of the preceding claims, characterized in that said change in said photochromic state of said FRET acceptor in at least one direction is caused by irradiation with ultraviolet light.

9. A method according to any of the preceding claims, characterized in that said change in said photochromic state of said FRET acceptor in at least one direction is caused by irradiation with visible light.

10. A method according to any of the preceding claims, characterized in that said excitation of said FRET acceptor is caused by irradiation with visible light.

11. A method according to any of the preceding claims, characterized in that the intensity of irradiation used to change said photochromic state of said FRET

acceptor is substantially stronger than the intensity of irradiation used to generate the observed fluorescence.

12. A method according to any of the preceding claims, characterized in that said sample is irradiated in a temporally modulated fashion in order to change said photochromic state of said FRET acceptor.

13. A method according claims 7 and 12, characterized in that said sample is irradiated with light of said first wavelength and said second wavelength in an alternating fashion in order to change said photochromic state of said FRET acceptor.

14. An apparatus for determining a characteristic kinetic quantity of a chemical reaction in a sample involving a plurality of chemical species, at least one of said species including at least one fluorophore, comprising a sample carrier, at least one controllable light source for spectrally and temporally controlled irradiation of said sample located on said sample carrier, at least one light detector suitable for time-resolved measurements in order to detect fluorescence light which is emitted from said sample due to said irradiation, and a control unit adapted to control said at least one light source and said at least one light detector according to the method of one of the preceding claims.

15. An apparatus according to claim 14, characterized in that an evaluation unit is provided for automated evaluation of detected fluorescence light data in order to calculate said characteristic kinetic quantity.

16. An apparatus according to one of the claims 14 or 15, characterized in that all components of the apparatus are integrated in a portable housing.